Reference: *Biol. Bull.* **169:** 565–577. (December, 1985)

CHANGES IN BEHAVIOR AND OCELLAR STRUCTURE DURING THE LARVAL LIFE OF SOLITARY ASCIDIANS

SHOGO KAJIWARA[1] AND MASAO YOSHIDA*[2]

[1]*Marine Biological Station, Tohoku University, Asamushi, Aomori 039-34, Japan, and* [2]*Ushimado Marine Laboratory, Kashino, Ushimado, Okayama 701-43, Japan*

ABSTRACT

Larvae of *Ciona savignyi* Herdman change swimming behavior during the course of development. Newly hatched larvae swim upward by negative geotaxis, accumulating beneath the water surface. Thirty minutes after hatching, the two-dimensional spread becomes more regionally restricted and the aggregated pattern looks like a swarm of mosquitoes. One and a half hours after hatching, larvae become photoresponsive, swimming upward for a short time immediately after shading. Meanwhile, the duration of swimming induced by shading becomes longer. Two and a half hours after hatching, larvae are weakly photonegative. Three and a half hours after hatching, swarming is abolished and photonegativity is much stronger than the previous stage.

The larval ocellus differentiates after the tadpole hatches. In newly hatched larvae, the flat pigment cell contains sparsely scattered pigment granules. Several short tubular membranes derived from ciliary endings of photoreceptor cells are irregularly arranged. One hour after hatching, the pigment cell becomes roughly L-shaped. The originally tubular membranes become paddle-shaped and increase in number and size. The pigment cell then assumes a V- or J-shape and becomes loaded with densely packed pigment granules. Some of the paddle-shaped membranes are arranged into lamellae and increase greatly in number and length. Three and a half hours after hatching, the ocellus becomes fully differentiated.

Using morphometrical parameters as regards the size of photoreceptor endings and the disposition of pigment granules, we show that changes in photic behavior coincides roughly with the course of differentiation of the ocellar elements.

INTRODUCTION

Larvae of sessile marine invertebrates are generally pelagic and respond to ecological factors in species-specific ways, by which they reach the substratum. One of the predominant ecological factors involved in larval settling is light (Thorson, 1964, for review). Although the morphology of the photoreceptor systems of invertebrates has been studied widely (Eakin, 1970, for review), works on the dynamic relationship between changes in photic behavior and photoreceptor morphogenesis during the larval life are rare (Young and Chia, 1982, in a polychaete; Chia and Koss, 1983, in an opisthobranch). In ascidians, several investigators (Grave, 1920; Mast, 1921; Woodbridge, 1924; Crisp and Ghobashy, 1971) explored larval behavior, while others (Dilly, 1961, 1964; Eakin and Kuda, 1971; Barnes, 1971) studied the ultrastructure of fully differentiated larval ocelli. The work of Barnes (1974) on "embryos" of *Amaroucium constellatum* is the only one to deal with the differentiation of the ocellus. Here we report that behavioral changes occurring during the course of solitary ascidian

Received 1 February 1985; accepted 13 September 1985.

* To whom all the correspondence and reprints requests should be made.

larval life roughly correspond in time to photoreceptor differentiation. A substantial part of the present work has appeared in abstract form (Kajiwara and Yoshida, 1983).

Materials and Methods

Specimens of *Ciona savignyi* Herdman (Hoshino and Nishikawa, 1985) were reared on flower pots which were hung from the pier of the Ushimado Marine Laboratory into the sea. Gravid individuals were maintained in running seawater at 18–23°C and continuously illuminated with two 20 w fluorescent lamps (50 cm high from animals) for 1–10 days to prevent uncontrolled spawning (Lambert and Brandt, 1967). Additional observations were made using *Ascidia sydneiensis sames* Oka. Unless specified, however, the following descriptions concern *C. savignyi.* In both cases, gametes were obtained surgically from gonoducts. Cross-fertilized eggs were maintained at room temperature (21°C) after several washes with a large volume of filtered seawater. Hatching occurred nearly synchronously about 14 and 16 h after fertilization in *C. savignyi* and *A. sydneiensis samea,* respectively. Larvae which were collected within 15 min after the first larva hatched are referred to as "newly hatched larvae."

Larval behavior was studied as follows. A transparent plastic trough, 8 cm square and 5 cm deep, was placed under ambient room light enhanced with window light, the intensity of which (1300 lux) was adjusted by window-blinds. One side of the trough was parallel to the window such that a photic gradient was created across the trough, decreasing from the window side toward the inner wall. This arrangement was advantageous for examining the presence or absence of shadow reflexes as well as positive or negative phototaxis, because simply placing a black plate on the window side not only reduced the light intensity to 150 lux but also reversed the existing photic gradient across the trough. This reduction will be called "shading."

Distribution patterns of larvae were photographed at 15 min intervals from 15 min through 5 h after hatching. To record horizontal and vertical distribution patterns of larvae, two cameras were triggered simultaneously from above as well as from the side of the trough. The side camera was parallel to the window and a strobe flash was shone from the side opposite the window. The side camera looks beneath the water surface, recording not only the side view of the trough but also the reflected images of larvae just below the water surface. However, such an artifact was assumed to be immaterial for assessing changes in the distribution pattern of larvae in the middle and the deeper levels, and hence the presence or absence of their upward movements.

The trough contained 3 cm of seawater containing 4–6 larvae per ml. After gentle stirring, a transparent lid was put on the trough to avoid any air-borne disturbance to the water surface and the seawater was left undisturbed for 14 min until the first recording ("Initial" in Figs. 1, 3–5) was made. Shading commenced 5 s after the initial recording, and the distribution patterns were recorded at 2, 5, 10, 15, and 30 s after shading. The seawater was then stirred and the lid replaced until the Initial of the next recording period.

Differentiation of the structures associated with the cerebral vesicle was followed electron microscopically. Larvae were fixed at room temperature for 2 h in 2.5% glutaraldehyde buffered to pH 7.4 by 0.1 mol sodium cacodylate containing 0.4 mol sucrose. The fixed tissues were washed by three 15-min changes of the same buffer, post-fixed for 1 h in 1% osmium tetroxide in the same buffer, dehydrated through a graded series of ethanol, penetrated with propylene oxide, and embedded finally in an epoxy resin (TAAB embedding resin). Thin sections cut on a Porter-Blum MT-2 ultramicrotome were stained with alcoholic uranyl acetate and lead citrate, and examined with a Hitachi H-500H electron microscope.

Using an image analysis system (MOP-Videoplan, Kontron Electronic Group), various morphological parameters of photoreceptor endings and pigment cells were measured on enlarged images of EM negatives.

Results

Larval behavior

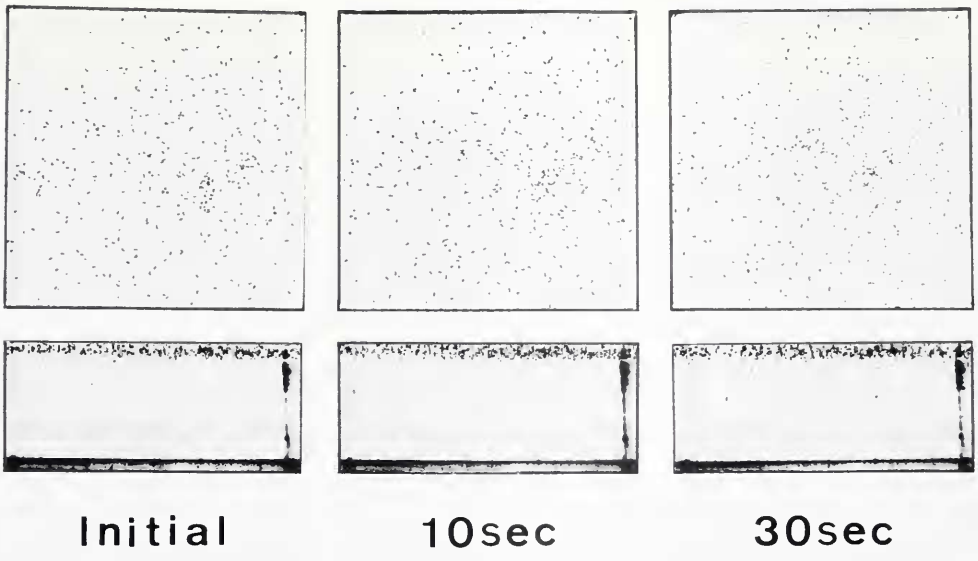
Using the behavioral pattern as a criterion, larval development was divided into five stages. Typical examples from 25 series of experiments are shown in Figures 1 through 5.

Stage I: until 15 min after hatching

Immediately after hatching, larvae move sporadically on the bottom of the trough by tail twiches. They soon begin to swim circularly at the substratum by brief bursts of tail beating. A few minutes later, the swimming increases in duration as well as in distance. In combination with negative geotaxis, larvae reach the water surface and swim circularly (Fig. 1). When observed from the front, these larvae appear to rotate clockwise with respect to their anterior-posterior axis. Stage I larvae of both *C. savignyi* and *A. sydneiensis samea* do not respond to changes in light conditions.

Stage II: Around 1 h after hatching

Larvae of both species are still indifferent to light. A notable phenomenon that begins to appear at this stage is the tendency to aggregate into a column which looks



FIGURES 1–5. Larval distribution patterns in a square trough photographed from above (upper row) and from a side (lower row), showing temporal sequence of changes of larvae at different stages. In Figures 1 and 3–5, shading was from the right side after initial recording and time after shading is given under each photograph. Scale: 4 cm.

FIGURE 1. The distribution pattern of Stage I larvae (15 min after hatching).

like a swarm of mosquitoes. This type of behavior starts 30 min after hatching in *C. savignyi* and 1 h in *A. sydneiensis samea.* The sequence of swarm formation is shown in Figure 2. Close observation of individual larvae revealed that while swimming beneath the water surface, they often stopped swimming and sank passively. The frequency of larval stopping was increased when two larvae collided with each other. After sinking to an indeterminate depth, larvae resumed upward swimming slightly outside the original sinking line. In this way, the larvae form first a ring pattern when viewed from above (2.5 min in Fig. 2) which becomes smaller and smaller until a single column is formed at the end (4.5 min in Fig. 2).

Stage III: 1.5 h after hatching

Larvae begin to show a shadow reflex, swimming upward within 2 s after shading. Note that in the side view of the 10 s recording in Figure 3, the number of larvae on the bottom is much reduced. Negative phototaxis is not apparent as yet, so that the distribution pattern in the trough remains the same as Stage II to the end of the recording period (30 s in Fig. 3).

Stage IV: 2.5 h after hatching

Larvae still form the swarm as before but now the size increases because the degree of aggregation has decreased (Initial in Fig. 4). A notable difference from the preceding stage is that the swarm disperses in about 10 s after shading. The massive shift towards the darker side (rightside in 10 s in Fig. 4) during shading indicates the onset of photonegativity. 30 s after shading, a few larvae begin to sink slowly. They form a swarm again after about 10 min when left undisturbed. Larvae of *A. sydneiensis samea* also become negatively phototactic about 3 h after hatching.

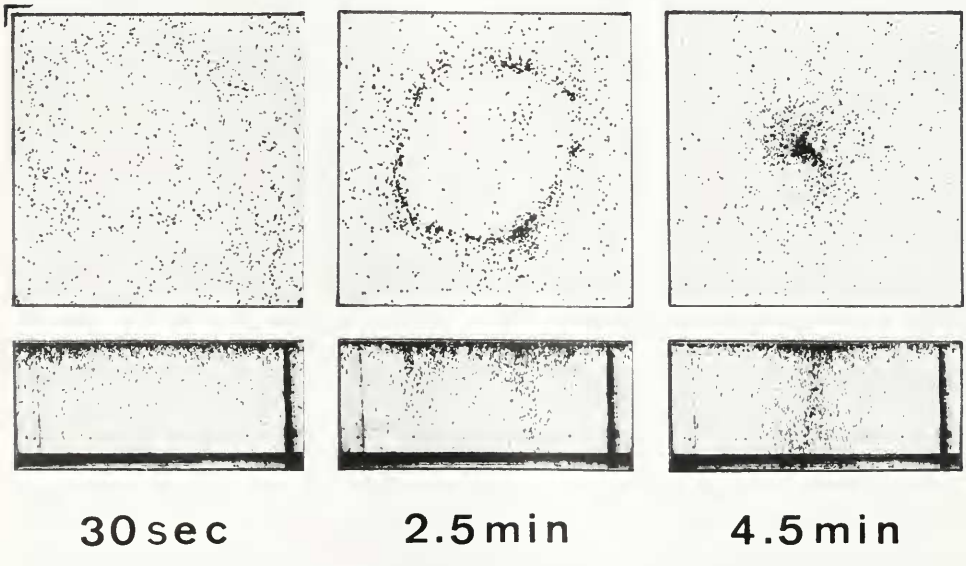


FIGURE 2. The sequence of swarm formation which occurs between Stage II and IV. This experiment was done with Stage III larvae. The time after stirring seawater is given under each photograph.

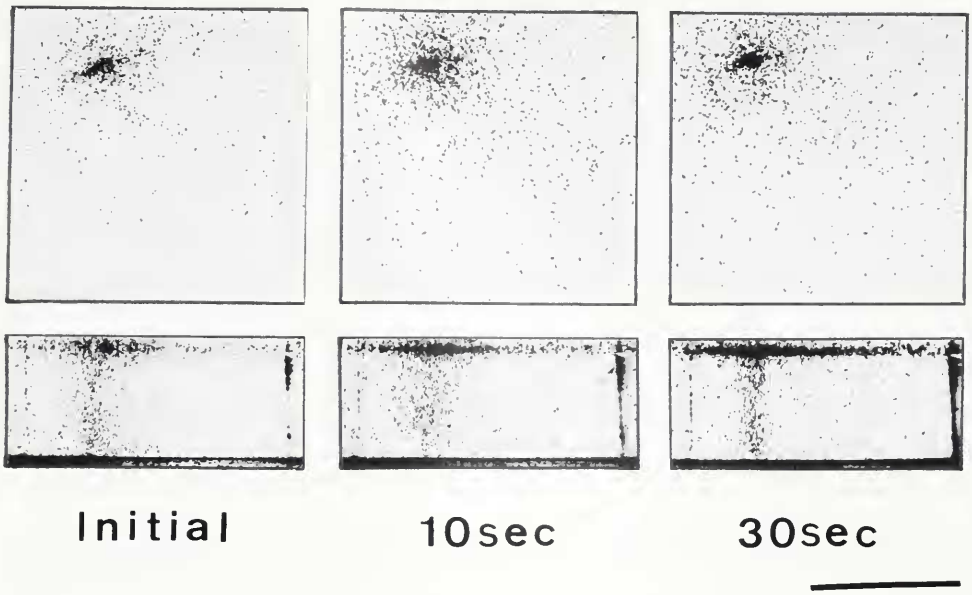


FIGURE 3. The distribution pattern of Stage III larvae (1.5 h after hatching).

Stage V: 3.5 h after hatching

As shown in the initial record in Figure 5, larvae no longer form the swarm but the majority are scattered on the left half, the side opposite the window due to the

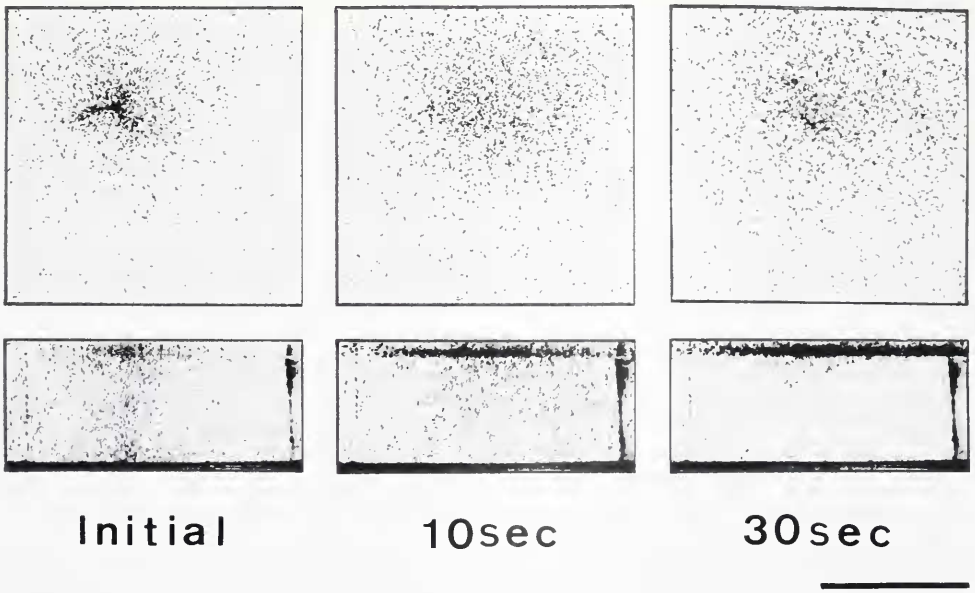


FIGURE 4. The distribution pattern of Stage IV larvae (2.5 h after hatching).

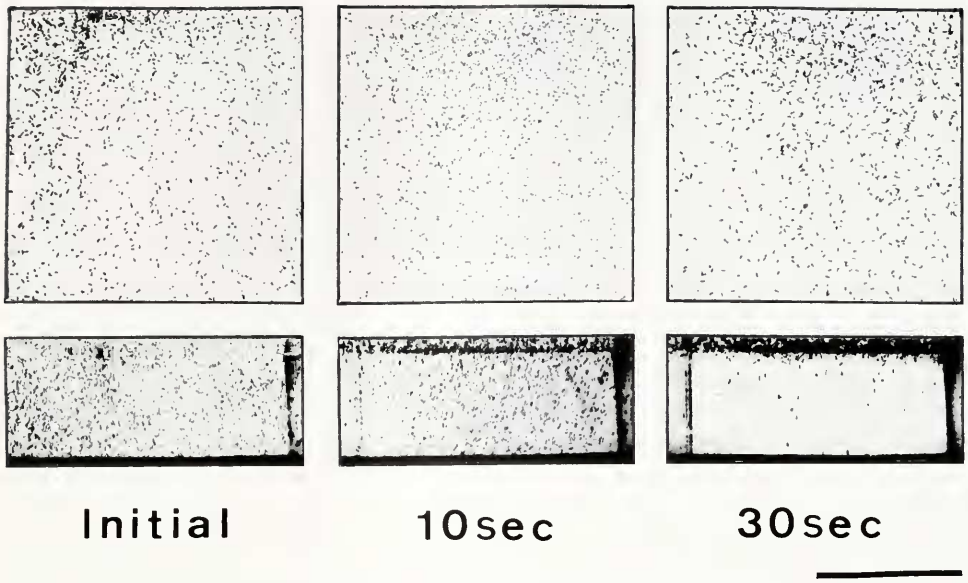


FIGURE 5. The distribution pattern of Stage V larvae (3.5 h after hatching).

photonegativity. Immediately after shading, larvae begin swimming upward as well as to the darker side (right side in 10 s in Fig. 5). Thus, the photonegative response is stronger than before. By 15 s after shading, the majority reach the water surface and a few begin to sink again 30 s after shading.

Differentiation of ocellus

The structure of the fully differentiated ocellus has been described by Dilly (1961, 1964), Eakin and Kuda (1971), and Barnes (1971), so that a brief description is sufficient here. In both species examined, the ocellus is located on the right-posterior wall of the cerebral vesicle, pointing ventro-laterally (Fig. 6a, b). Each ocellus consists of three components (Fig. 6c). (1) One cup-shaped pigment cell which is loaded with membrane-bounded melanin granules (Whittaker, 1973, 1979) appears as a whole mass, V-, or J-shaped in cross sections. (2) About 15–20 photoreceptor cells line up on the lateral side of the pigment cell. Each cell extends a narrow process through the pigment cell giving rise to about 25 lamellae from the ciliary projection within the cup lumen. (3) Three lens cells are arranged in a row and each of them contains a large lens vesicle bordered by mitochondria.

To investigate whether there are any changes in the morphological differentiation of the ocellus corresponding to changes in larval behavior, materials were fixed at the 5 stages defined by behavioral criteria. Morphometric analysis included the following five parameters, using transverse profiles of five ocelli for each stage. For photoreceptive elements in each ocellus, the cell whose lamellae were greatest in number and cut longitudinally was chosen. In this cell the number of lamellae was counted and the length of these lamellae was measured. For pigment cells, the cell profile showing the largest cup lumen was chosen and the following determined: (1) size of pigment granules, (2) ratio of the total area occupied by pigment granules to that of the pigment cell (nuclear zone excluded), and (3) ratio of the total area occupied by pigment granules

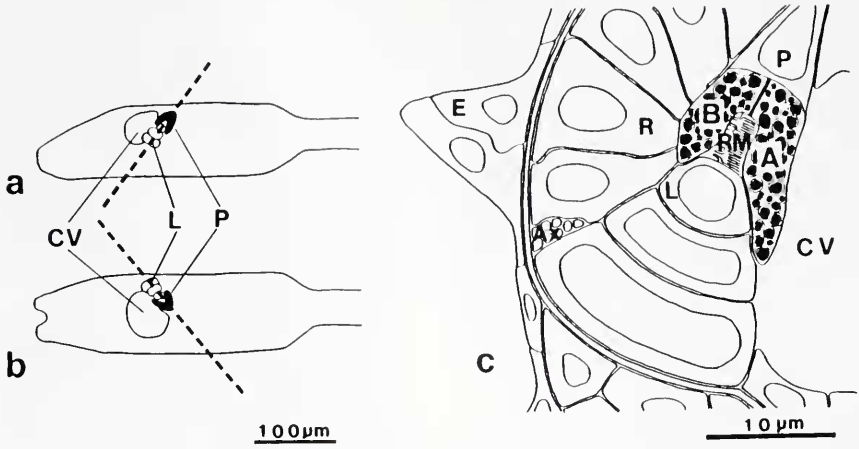


FIGURE 6. Semi-diagrammatic representation (roughly to scale) of the gross appearance of the ocellus, constructed from serial LM and EM sections. a and b: Lateral and horizontal profiles, respectively. c: An expected profile when the plane of the section passes the oblique broken lines drawn in a and b. Ax: axons of photoreceptor cells, CV: cerebral vesicle, E: epidermal cell, L: lens cell, P: pigment cell, R: photoreceptor cell, RM: photoreceptive membranes.

in the lateral side of the cup (B in Fig. 6c) to that in the medial side (A in the same Figure).

Stage I: until 15 min after hatching

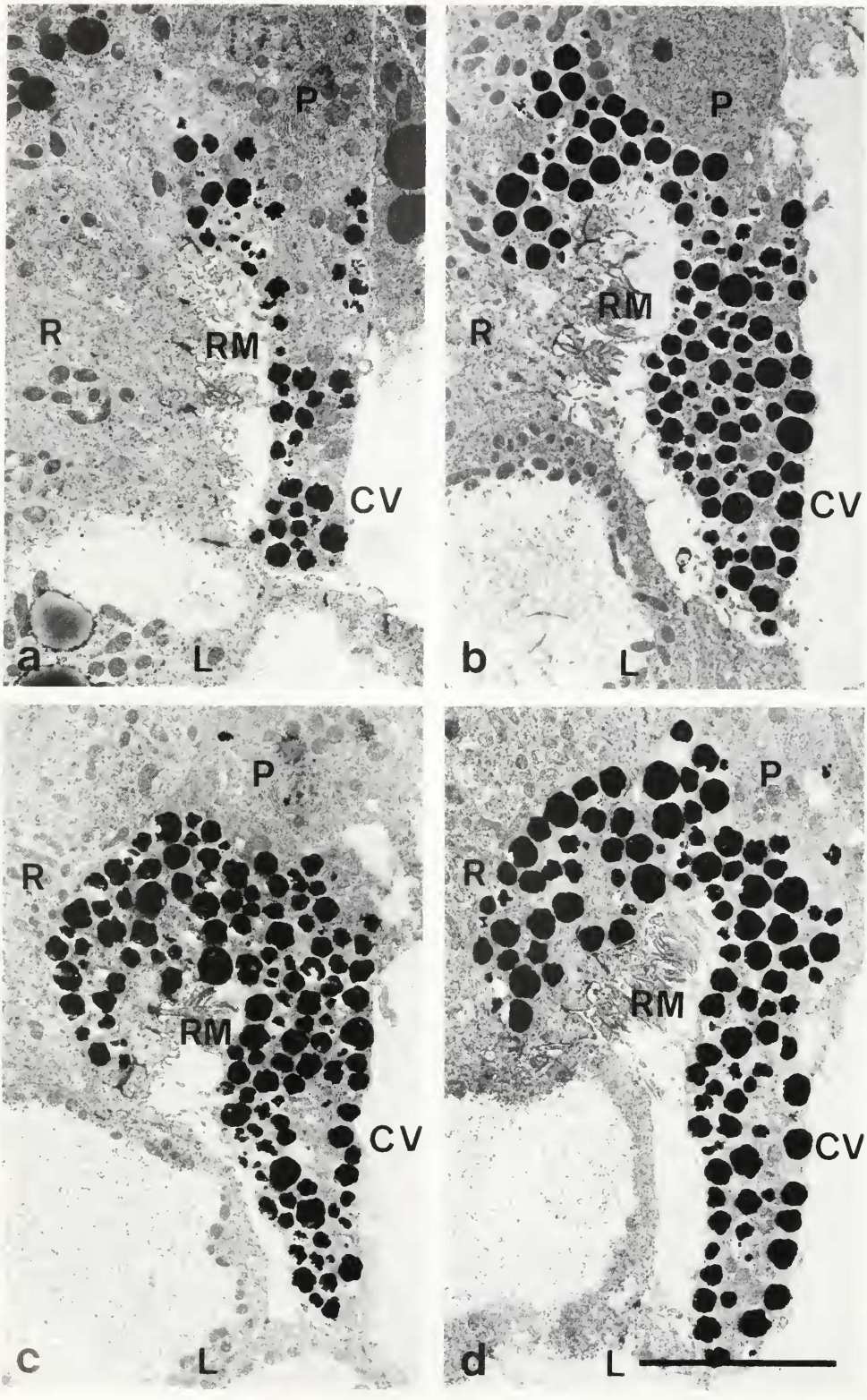
The pigment cell does not extend the B-portion in Figure 6c (Fig. 7a). The pigment granules are small in size (0.15 ± 0.04 μm; n = 382, Fig. 10b) and in number so that the area covered by the pigment granules occupies 31.3 ± 6.9% of the pigment cell cytoplasm. Some of the membranes derived from the apex of the photoreceptor cell are tubular as revealed by the cross-section in Figure 9a. The number of lamellae arising from single cells ranges from 9 to 16. They are short (0.47 ± 0.07 μm; n = 60, Fig. 10a) and irregularly oriented (Fig. 8a).

Stage II: 1 h after hatching

The pigmented area starts to extend toward the lateral side (B-portion in Fig. 6c) of the cerebral ganglion. Pigment granules become larger (0.19 ± 0.02 μm in diameter; n = 483, Fig. 10b) and occupy a larger portion of the pigment cell (42.3 ± 5.8%, Fig. 10b). The lamellae from the photoreceptor cell increase in number, ranging from 16 to 28, from single cells and in length (0.58 ± 0.06 μm; n = 92, Fig. 10a).

Stage III: 1.5 h after hatching

The growth of the B-portion of the pigment cell continues so that the pigmented area now appears roughly J-shaped (Fig. 7b). Pigment granules become larger (0.26 ± 0.04 μm in diameter; n = 379, Fig. 10b). Membranes arising from the photoreceptor projections are transformed into a paddle-shape (Fig. 9b). The number of lamellae from single cells ranged from 19 to 23 and the average length was 0.62 ± 0.07 μm (n = 109, Fig. 10a).


P
R
RM
CV
a
L
P
R
RM
CV
b
L
P
R
RM
CV
c
L
P
R
RM
CV
d
L

Stage IV: 2.5 h after hatching

The extension of the B-portion nears completion so that the pigmented area takes a V-shape form. At this stage, narrow processes of the photoreceptor cells are surrounded by the pigment cell. As shown in Figure 10b, the average diameter of the pigment granules (0.24 ± 0.02 μm; n = 672) and the ratio of the pigmented area to the cytoplasm (48.3 ± 2.6%) are not much different from the previous stage, but the ratio of the pigmented area in B to that in A is markedly increased, from 26.4 ± 4.4% in Stage III to 55.5 ± 11.0% in Stage IV (Fig. 10b). It is also noteworthy that although the number of lamellae arising from single cells is not much increased (range: from 20 to 28), their length (0.88 ± 0.07 μm; n = 120) are approaching the maximum value in the next stage (Fig. 10a).

Stage V: 3.5 hours after hatching

The larval eye in this stage (Fig. 7d, Fig. 8d) is fully differentiated. Morphometrical values of all five parameters fall within the standard deviation of those in the previous stage (Fig. 10a, b).

Discussion

Sessil marine invertebrates are found in restricted zones of rocks or sand beaches. Pelagic larvae disperse themselves and finally choose the best-fitted substratum for future life. This adaptive strategy is achieved through dynamic changes in response to various ecological factors. The temporal sequence of changes in the larval behavior of the ascidian, *C. savignyi,* may be summarized as follows (refer to Fig. 10a, inset). (1) Newly hatched larvae swim up geonegatively and are indifferent to light. (2) Thirty min after hatching, larvae begin to form a swarm under all light conditions. (3) One and a half h after hatching, shadow reflexes that last until metamorphosis begins to appear. (4) Two and a half h after hatching, larvae become negatively phototactic though still weak. (5) Three and a half h after hatching, larvae no longer form the swarm and the shadow reflex and photonegative response are strongest. Metamorphosis occurs at any time between 5 h and 2 weeks after hatching.

From these behavioral patterns in the experimental trough, we can predict the larval behavior in the field: newly hatched larvae swim up to the water surface and are dispersed with water currents as they repeat up and down excursions in the upper stratum. Later, larvae become photonegative and occasionally stop swimming, sinking down to a deeper layer. Upon reduction in light intensity, they swim toward a darker area as well as upward. In this way, they will tend to settle on under-surfaces of the substrata in their late stages.

The behavioral patterns described above appear to be typical of solitary ascidians. Similar behaviors were also described in *Styela partita* (Grave, 1941, 1944). However, the larvae of many compound ascidians show positive phototaxis for a short period in the early stage, and soon it changes to negative phototaxis (*Amaroucium constellatum:* Grave, 1920, Mast, 1921; *Amaroucium pellucidum:* Mast, 1921; *Botryllus schlosseri:* Woobridge, 1924; *Diplosoma listerianum:* Crisp and Ghobashy, 1971).

It may be expected that the functional differentiation of ocelli would correspond

---

FIGURE 7. Changes in the shape of pigmented area during larval life. a: Stage I, b: Stage III, c: Stage IV, d: Stage V. CV: cerebral vesicle, L: lens cell, P: pigment cell, R: photoreceptor cell, RM: photoreceptive membranes. Scale 5 μm.

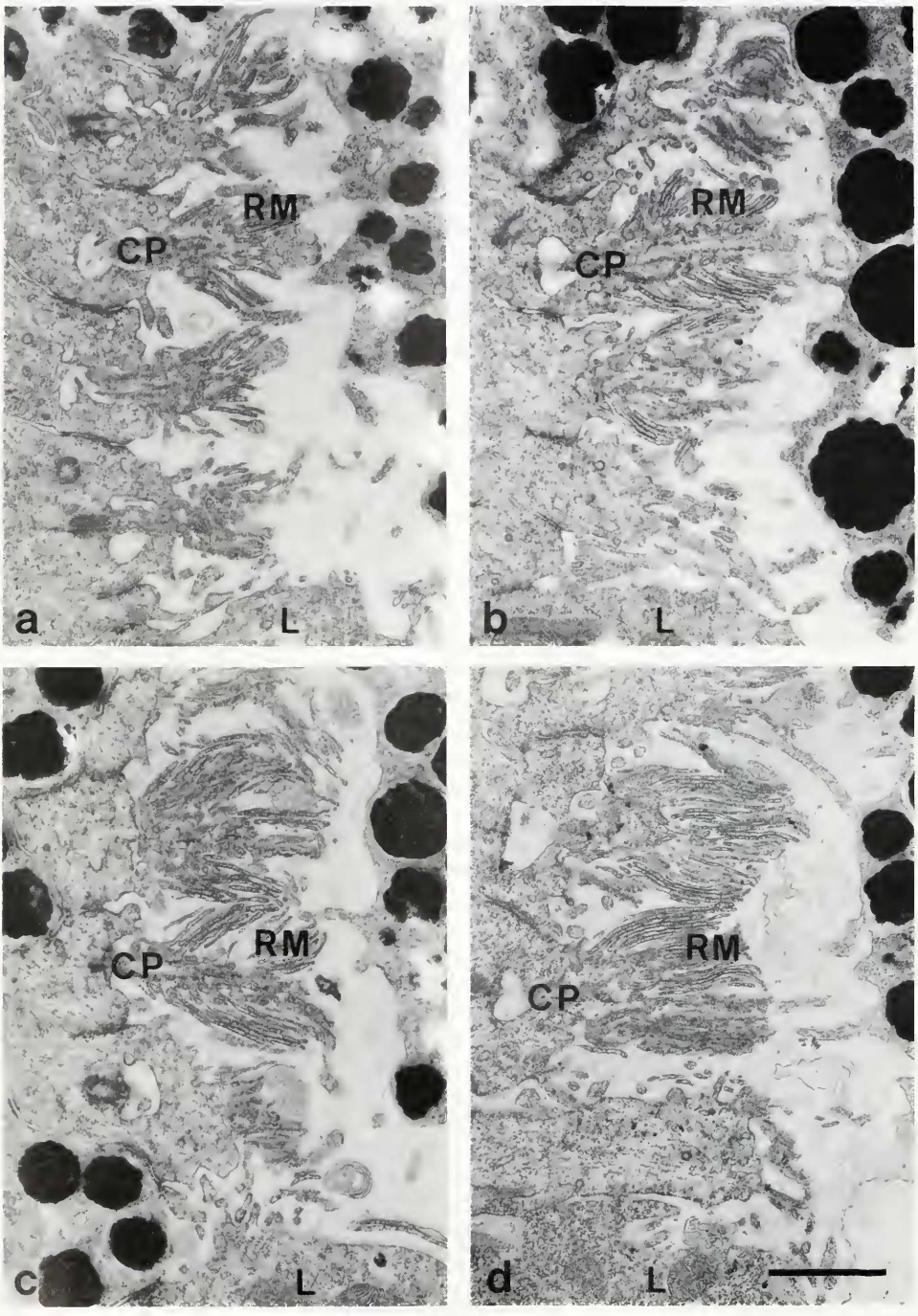


FIGURE 8. Changes in the ultrastructure of the photoreceptive membranes during larval life. a: Stage I, b: Stage III, c: Stage IV, d: Stage V. CP: ciliary projection, L: lense cell, RM: photoreceptive membranes. Scale: 1 μm.

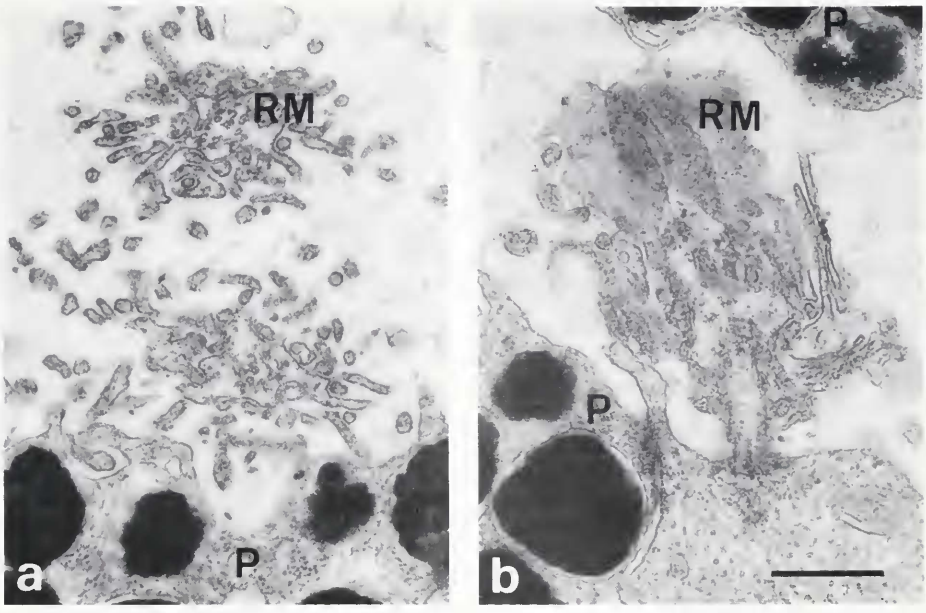


FIGURE 9. Magnified profiles of the distal part of photoreceptor cell. a: Stage I. b: Stage III. P: pigment cell, RM: photoreceptive membranes. Scale: 0.5 μm.

to the development of photic responses. Although the ultrastructure of ocelli has been studied widely in differentiated larvae (Dilly, 1961, 1964; Barnes, 1971; Eakin and Kuda, 1971), the work of Barnes (1974) on "embryos" of *Amaroucium constellatum*, a compound ascidian, was the only one that followed the differentiation of the ocellus. Our observations confirmed and extended those of Barnes by introducing morphometrical aspects. Differentiation of photoreceptive membranes and the pigment cell proceeds rapidly within 3 h. One and a half h after hatching (Stage III) the larvae become responsive to a shadow. Although we could not ascertain the number of newly recruited photoreceptor cells between Stage I and Stage III, the increase in surface area of the photoreceptive membrane as estimated by the number of lamellae from single cells (from 12.0 to 21.8) and their length (from 0.47 to 0.62 μm) should account for the functional maturation of the presumptive photoreceptor cells.

The negative phototaxis then appearing from Stage IV onward is accompanied by a more than two fold increase in the ratio of the pigmented area in the B-portion to that in A-portion, resulting in both of the lateral sides of the ocellar lumen becoming loaded with pigment granules. This pigment disposition may aid the photoreceptors in sensing the direction of the light source. The arrangement of lamellae perpendicular to the incident light coming through the lens system may also be advantageous for photosensitivity. Considering that ocelli of newly hatched larvae which are indifferent to light are already equipped with the three major components (a single pigment cell, photoreceptor cells and three lens cells) of the ocellus, maturation of the central nervous network may also be needed for performance of complex behavioral responses.

Although swarm formation may only be an experimental phenomenon, its appearance and disappearance could be taken as a manifestation of dynamic changes in the developing nervous mechanisms at the middle of larval life. For the swarms to be formed, mechanoreceptors such as the pressure receptors in the cerebral vesicle

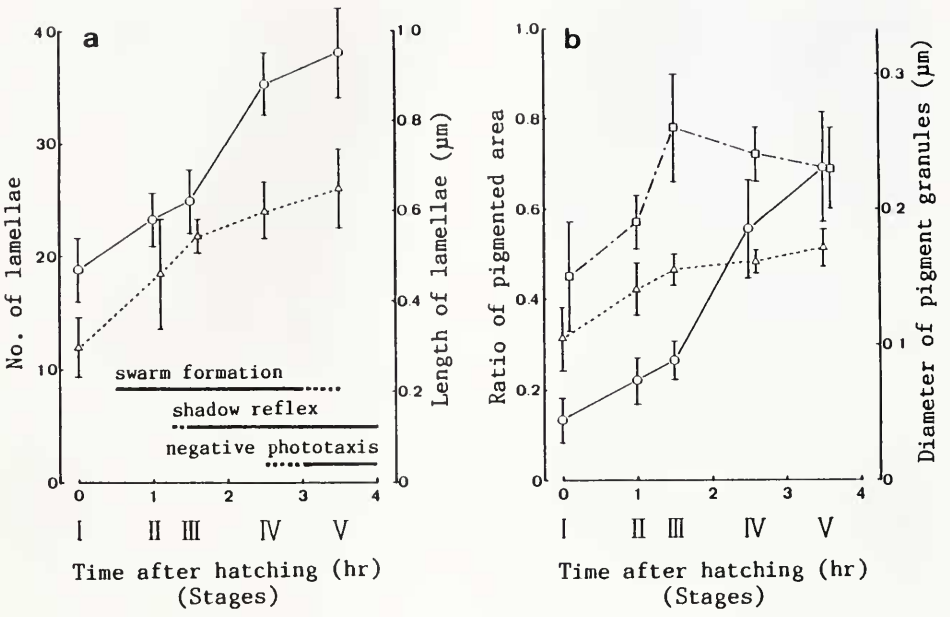


FIGURE 10. Changes in morphometric parameters of photoreceptive membranes (a) and pigment granules (b) during larval life (abscissae: time after hatching in h and corresponding stages). a: Triangles show the number of lamellae arising from single cells (ordinate on the left) and circles, the length of lamellae (ordinate on the right). Inset shows the temporal sequence of the appearance and disappearance of three behavioral criteria (dots: weak responses). b: Squares show the diameter of pigment granules in cross-sections (ordinate on the right), triangles, the ratio of the pigmented area to the cytoplasma and circles, the ratio of the pigmented area in the B-portion in Figure 6c to that in the A-portion (for the latter two, ordinate on the left).

(Eakin and Kuda, 1971; Reverberi, 1979) and sensory cilia in the tail epidermis (Torrence and Cloney, 1982) could be involved in sensing the depth and the contact with neighboring larvae, respectively. The fact that the swarm disappears as metamorphosis approaches may suggest that an integrating mechanism which has been concerned with mechanical stimuli during the middle of larval life may be taken over by photosensory mechanisms which will be important for seeking a substratum to settle.

The sensory and neural mechanisms involved in larval behaviors of solitary and compound ascidians remains to be studied in more detail.

ACKNOWLEDGMENTS

We thank Professor C. C. Lambert, Department of Biological Science, California State University, for his comments on the manuscript, Dr. T. Numakunai, Asamushi Marine Laboratory, for his aid in taxonomy of the materials used, and Mr. M. Isozaki and Mr. W. Godo for their technical help. The work was supported in part by a grant-in-aid to M.Y. from the Ministry of Education, Science and Culture of Japan.

LITERATURE CITED

BARNES, S. N. 1971. Fine structure of the photoreceptor and cerebral ganglion of the tadpole larva of *Amaroucium constellatum* (Verrill). (Subphylum: Urochordata; Class: Ascidiacea). *Z. Zellforsch.* **117**: 1–16.

BARNES, S. N. 1974. Fine structure of the photoreceptor of the ascidian tadpole during development. *Cell Tissue Res.* **155**: 27–45.

CHIA, F.-S., AND R. KOSS. 1983. Fine structure of the larval eyes of *Rostanga pulchra* (Mollusca, Opisthobranchia, Nudibranchia). *Zoomorphology* **102**: 1–10.

CRISP, D. J., AND A. F. A. A. GHOBASHY. 1971. Responses of the larvae of *Diplosoma listerianum* to light and gravity. Pp. 443–465 in *Fourth European Marine Biology Symposium*, D. J. Crisp, ed. Cambridge University Press, Cambridge.

DILLY, N. 1961. Electron microscope observations of the receptors in the sensory vesicle of the ascidian tadpole. *Nature* **191**: 786–787.

DILLY, N. 1964. Studies on the receptors in the cerebral vesicle of the ascidian tadpole. 2. The ocellus. *Q. J. Microsc. Sci.* **105**: 13–20.

EAKIN, R. M. 1970. Structure of invertebrate photoreceptors. Pp. 625–684 in *Handbook of Sensory Physiology, Vol. VII/1, Photochemistry of Vision*, H. J. Dartnall, ed. Springer-Verlag, Berlin-Heidelberg-New York.

EAKIN, R. M., AND A. KUDA. 1971. Ultrastructure of sensory receptors in ascidian tadpoles. *A. Zellforsch.* **112**: 287–312.

GRAVE, C. 1920. *Amaroucium pellucidum* (Leidy) form *constellatum* (Verrill). I. The activities and reactions of the tadpole larva. *J. Exp. Zool.* **30**: 238–257.

GRAVE, C. 1941. The "eye-spot" and light-responses of the larvae of *Cynthia partita*. *Biol. Bull.* **81**: 287.

GRAVE, C. 1944. The larva of *Styela (Cynthia) partita:* Structure, activities and duration of life. *J. Morphol.* **75**: 173–191.

HOSHINO, Z., AND T. NISHIKAWA. 1985. Taxonomic studies of *Ciona intestinalis* (L.) and its allies. *Publ. Seto Mar. Biol. Lab.* **30**: 61–79.

KAJIWARA, S., AND M. YOSHIDA. 1983. Changes in behavior and photoreceptoral architecture of an ascidian tadpole during the course of development. Pp. 68 in *Proceedings of the Fifth Annual Meeting of the Japan Society for General and Comparative Physiology.*

LAMBERT, C. C., AND C. L. BRANDT. 1967. The effect of light on the spawning of *Ciona intestinalis*. *Biol. Bull.* **132**: 222–228.

MAST, S. O. 1921. Reactions to light in the larvae of the ascidians, *Amaroucium constellatum* and *Amaroucium pellucidum* with special reference to photic orientation. *J. Exp. Zool.* **34**: 148–187.

REVERBERI, G. 1979. On the third sensorial organ of ascidian tadpoles. *Acta Embryol. Exp.* **1**: 91–99.

THORSON, G. 1964. Light as an ecological factor in the dispersal and settlement of larvae of marine bottom invertebrates. *Ophelia* **1**: 167–208.

TORRENCE, S. A., AND R. A. CLONEY. 1982. Nervous system of ascidian larvae: caudal primary sensory neurons. *Zoomorphology* **99**: 103–115.

WHITTAKER, J. R. 1973. Tyrosinase in the presumptive pigment cells of ascidian embryos: tyrosine accessibility may initiate melanin synthesis. *Dev. Biol.* **30**: 441–454.

WHITTAKER, J. R. 1979. Quantitative control of end products in the melanocyte lineage of the ascidian embryo. *Dev. Biol.* **73**: 76–83.

WOODBRIDGE, H. 1924. *Botryllus schlosseri* (Pallas): the behavior of the larva with special reference to the habitat. *Biol. Bull.* **47**: 223–230.

YOUNG, C. M., AND F.-S. CHIA. 1982. Ontogeny of phototaxis during larval development of the sedentary polychaete, *Serpula vermicularis* (L.). *Biol. Bull.* **162**: 457–468.